Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

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Letter to the Editor

Detection of a SARS-CoV-2 P.1.1 variant lacking N501Y in a vaccinated health care worker in Italy

Dear Editor,

We read with interest the recently published manuscript of Dimeglio et al., exploring the SARS-CoV-2 immune response and vaccination of healthcare workers post-infection.[1] and here we report a case of SARS-CoV-2 infection with a P.1.1 variant lacking the Y501 mutation in a vaccinated individual in Italy.

COVID-19 vaccines are very effective in preventing infections, hospitalizations and deaths. However, several cases of SARS-CoV-2 infections have been reported in vaccinated individuals (called "vaccine breakthrough cases") inoculated with one or both doses of vaccine.[2] This indicates that a small percentage of fully vaccinated subjects can be infected when exposed to the virus. Recently, in the UK, South Africa, Brazil and most recently in India.[3–5] SARS-CoV-2 variants of concern (VOC) have been identified. These new variants harbor mutations in the spike protein, and particularly in the receptor binding domain (RBD). These VOC are important because they show that a number of viral mutations are emerging with a potential impact on infectivity, immune escape and vaccine effectiveness.

In the University Campus Biomedico Hospital (UCMB – Rome, Italy), vaccination of health care workers began on December, 2020 soon after followed by active monitoring of potential breakthrough cases. For this reason, all health care workers, regardless of their symptomatic status, were tested weekly by molecular assay on nasopharyngeal swabs.

Here, we report the first case of SARS-CoV-2 P.1.1 infection lacking N501 mutation in a fully vaccinated (Pfizer) 22-year-old female nurse, working in the COVID-Center of the UCBM.

In early-January 2021, the nurse received the first vaccine dose followed by the second shot three weeks after. Two weeks after second dose of vaccine, a first Quantitative IgG anti-spike chemiluminescent assay tested positive with 2362 BAU/mL. Quantitative IgG anti-spike assay was repeated after 45 and 90 days confirming positive results with 1029 BAU/mL and 432 BAU/mL, respectively. Three months after the second dose the patient started presenting mild symptoms (headache and fever), compatible with a viral infection. At this time, a rapid SARS-CoV-2 N protein Chemiluminescent assay was performed on nasopharyngeal swab, revealing a positive result (> 5000 pg/mL). The same swab was then tested for the detection of SARS-CoV-2 RNA by multiplex real-time PCR Allplex™ SARS-CoV-2 assay (Seegene Inc, Seoul, Korea). Cycle threshold values (Cts) of N, E and RdRp/S targets were 17, 18, and 18, respectively. Whole genome sequencing was then conducted on the same swab by Myseq II Illumina. Consensus sequences were generated by de novo assembling using Genome Detective (https://www.genomedetective.com/).[6] A total of 1,323,357 mapped reads were obtained, resulting in a sequencing mean depth > 1,000X and a coverage of > 99.8%. Sequences were aligned using MAFFT[7] and submitted to IQ-TREE 2 for maximum likelihood (ML) phylogenetic analysis.[8]

Lineages assessment, conducted using Phylogenetic Assignment of Named Global Outbreak Lineages tool (available at https://github.com/hCoV-2019/pangolin), revealed the new strain belonged to the P.1.1 lineage. Phylogenetic inference by combining our new isolate (EPI_ISL_2,488,760) with a representative dataset available on GISAID (https://www.gisaid.org/) up to May 31th, 2021 demonstrated that the newly obtained genome belongs to P.1.1 lineage and clustered significantly with SARS-CoV-2 P.1.1 strains isolated in Italy between March and May 2021 (Fig. 1a, b) (Bootstrap=0.80, SH-aLTR=0.80). Further, we analyzed the mutational profile of the newly generated strain to determine its lineage-defining mutations. The identified lineage harbored all the P.1.1 lineage specific mutations (Fig. 1c), with the exception of the N501Y, a specific mutation that seems to be linked with an increased transmissibility.[9] We also identified another Spike mutation, namely S640F, which currently is growing in many samples worldwide[10] (Fig, 1c).

As observed through the course of the pandemic, these mutations highlight the ability of SARS-CoV-2 to generate new viral strains. In particular, these newly identified mutations likely represent a new SARS-CoV-2 variant probably able to escape vaccine protection, although to a certain extent, as indicated by the lack of severe symptoms. The lack of the N501Y may be linked to high transmissibility and immune evasion as indicated also for the new path followed by the delta new variant (B1.617.2). Probably, the vaccine immune pressure could select less prevalent strains leading to their emergence.

At present, it is not clear how widely this variant is currently present in Italy and worldwide. Nonetheless, our case report, clearly indicate the importance of genetic sequencing and analysis to promptly identify and characterize variants as soon as possible both in vaccinated individuals and in the general population.

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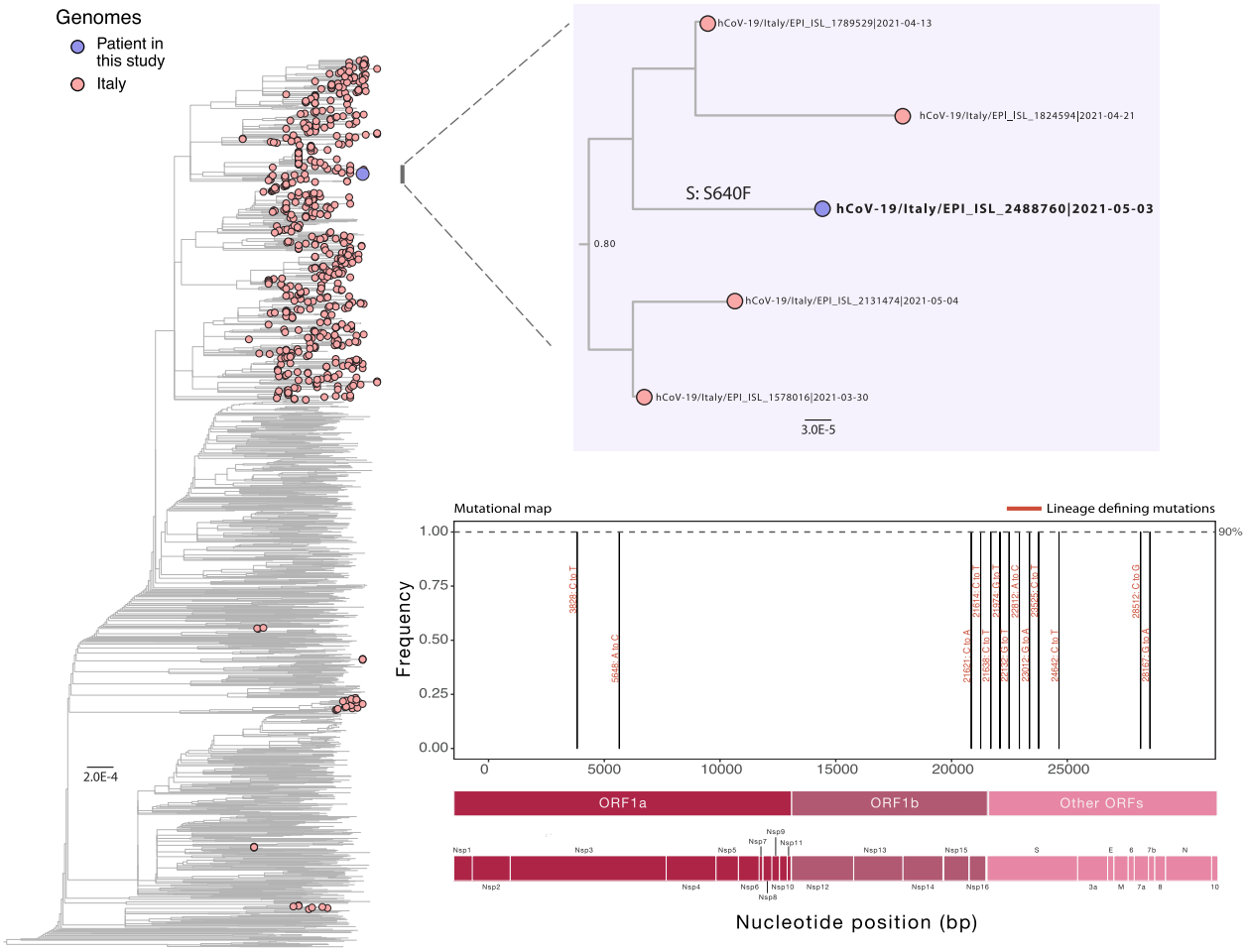

Fig. 1. Genomic detection of the SARS-CoV-2 P.1 variant of concern in a vaccine breakthrough case in Italy. (a) Maximum likelihood (ML) phylogenetic tree including the newly isolate obtained in this study plus n=1631 SARS-CoV-2 strains belonging to the P.1 lineage collected up to May 28th, 2021. (b) Representation of the zoom of the Italian P.1 clade. Branch support (Bootstrap = 0.80, SH-aLTR = 0.80) is shown at key node. c) Variant map of the P.1 lineage-defining-mutations was mapped against the SARS-CoV-2 genome structure. Lineage-defining mutation are highlighted in red (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.).

Ethical statement

This research was approved by the University of Campus Biomedico Ethics Review Committee (Approval number 8.1(21).21 OSS).

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Declaration of Competing Interest

The authors declare no competing interests.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.jinf.2021.06.026.

References

1. Dimeglio C, Herin F, Miedougé M, Da-Silva I, Porcheron M, Martin-Blondel G, Soulat JM, Izopet J. One year later: SARS-CoV-2 immune response and vaccination of healthcare workers post-infection. *J Infect* 2021 Jun 17 S0163-4453(21)00312-1Epub ahead of print. PMID: 34147528. doi:10.1016/j.jinf.2021.06.016.

2. Hacisuleyman E, Hale C, Saito Y, Blachere NE, Bergh M, Conlon EG, Schaefer-Babajew DJ, DaSilva J, Muecksch F, Gaebler C, Lifton R, Nussenzweig MC, Hatziioannou T, Bieniasz PD, Darnell RB. Vaccine breakthrough infections with SARS-CoV-2 variants. *N Engl J Med* 2021;**384**(23):2212–18 Jun 10Epub 2021 Apr 21. PMID: 33882219; PMCID: PMC8117968. doi:10.1056/NEJMoa2105000.
3. Rambaut A, Loman N, Pybus O, et al. on behalf of COVID-19 Genomics Consortium UK (CoG-UK). Preliminary genomic characterisation of an emergent SARS-CoV-2 lineage in the UK defined by a novel set of spike mutations. https://virological.org/t/preliminary-genomic-characterisation-of-an-emergent-sars-cov-2-lineage-in-the-uk-defined-by-a-novel-set-of-spike-mutations/563. (accessed Dec 21, 2020).
4. Tegally H, Wilkinson E, Giovanetti M, et al. Detection of a SARS-CoV-2 variant of concern in South Africa. *Nature* 2021;**592**:438–43. doi:10.1038/s41586-021-03402-9.
5. Faria NR, Mellan TA, Whittaker C, et al. Genomics and epidemiology of the P.1 SARS-CoV-2 lineage in Manaus. *Braz Sci* 2021 Apr 14:eabh2644 Epub ahead of print. PMID: 33853970. doi:10.1126/science.abh2644.
6. Cleemput S, Dumon W, Fonseca V, Abdool Karim W, Giovanetti M, Alcantara LC, Deforche K, de Oliveira T. Genome detective coronavirus typing tool for rapid identification and characterization of novel coronavirus genomes. *Bioinformatics* 2020;**36**(11):3552–5 Jun 1PMID: 32108862; PMCID: PMC7112083. doi:10.1093/bioinformatics/btaa145.
7. Nakamura T, Yamada KD, Tomii K, Katoh K. Parallelization of MAFFT for large-scale multiple sequence alignments. *Bioinformatics* 2018;**34**:2490–2.
8. Minh BQ, Schmidt HA, Chernomor O, Schrempf D, Woodhams MD, von Haeseler A, Lanfear R. IQ-TREE 2: New models and efficient methods for phylogenetic inference in the genomic Era. *Mol Biol Evol* 2020;**37**:1530–4. doi:10.1093/molbev/msaa015 Medline.
9. Davies NG, Abbott S, Barnard RC, Jarvis CI, Kucharski AJ, Munday JD, Pearson CAB, Russell TW, Tully DC, Washburne AD, Wenseleers T, Gimma A, Waites W, Wong KLM, van Zandvoort K, Silverman JD, CMMID COVID-19 Working Group, COVID-19 Genomics UK (COG-UK) Consortium, Diaz-Ordaz K, Keogh R, Eggo RM, Funk S, Jit M, Atkins KE, Edmunds WJ. Estimated transmissibility and impact of SARS-CoV-2 lineage B.1.1.7 in England. *Science* 2021 Apr 9;**372**(6538):eabg3055 Epub 2021 Mar 3. PMID: 33658326; PMCID: PMC8128288. doi:10.1126/science.abg3055.
10. COG-UK-Consortium, 2021. (Available at: https://www.cogconsortium.uk/wp-content/uploads/2021/01/Report-2_COG-UK_SARS-CoV-2-Mutations.pdf)

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